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IDENTIFICATION OF THE LARVAL AGGREGATION PHEROMONE OF CODLING MOTH, *Cydia pomonella*

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Abstract—Mature larvae of the codling moth, *Cydia pomonella* L. (Lepidoptera: Olethreutidae), exit the fruit and seek sites suitable for pupation. Spinning cocoons in such sites, larvae produce a complex, cocoon-derived blend of volatiles recently shown to attract and/or arrest both conspecific larvae and the prepupal parasitoid *Mastrus ridibundus* Gravenhorst (Hymenoptera: Ichneumonidae). Here we report components of this blend that constitute the pheromone of fifth-instar *C. pomonella* larvae. Thirty-one two-choice olfactometer experiments showed that a blend of synthetic (*E*)-2-octenal, (*E*)-2-nonenal, sulcatone, and geranylacetone, in combination with either 3-carene and/or three saturated aldehydes (octanal, nonanal, decanal), elicited behavioral responses from *C. pomonella* larvae. In on-tree experiments with corrugated cardboard bands as pupation sites for larvae affixed to tree trunks, and with laboratory-reared larvae released onto such trees, more larvae cocooned in those halves of cardboard bands baited with cocoon-spinning conspecific larvae, or with synthetic pheromone components, than in unbaited control halves of the bands. With the larval aggregation pheromone identified in this study, there might be an opportunity to manipulate *C. pomonella* larvae in commercial fruit or nut orchards.

Key Words V Codling moth, larvae, *Cydia pomonella*, *Mastrus ridibundus*, aggregation pheromone, heptanal, octanal, nonanal, decanal, (*E*)-2-octenal, (*E*)-2-nonenal, sulcatone, geranylacetone, (+)-limonene, myrcene, 3-carene.

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INTRODUCTION

When mature larvae of the codling moth, *Cydia pomonella* L. (Lepidoptera: Olethreutidae), complete their development, they exit the fruit and seek sites suitable for pupation. While spinning cocoons in such sites, larvae produce an aggregation pheromone that attracts or arrests conspecific larvae (Duthie et al., 2003; Jumean et al., 2004a). Aggregation of fifth-instars prior to pupation may be part of a mating strategy (Duthie et al., 2003) because in laboratory bioassays, eclosed adult males appeared to be arrested by the sex pheromone (*E*,*E*)-8,10-dodecadienol emanating from mature female pupae, which may allow mating as soon as a female ecloses.

Identification of the cocoon-derived pheromone proved challenging because larval antennae were too small to be used effectively in gas chromatographicYelectroantennographic detection (GCYEAD) analyses of cocoon volatiles. Testing the hypothesis that *Mastrus ridibundus* Gravenhorst (Hymenoptera: Ichneumonidae), a parasitoid of late instar/prepupal *C. pomonella*, exploits odors produced by or associated with larvae as a kairomone during host-foraging, Jumean et al. (2004b) demonstrated (1) that 10 cocoon volatiles [3-carene, myrcene, heptanal, octanal, nonanal, decanal, (*E*)-2-octenal, (*E*)-2-nonenal, sulcatone, and geranylacetone] elicited responses from female *M. ridibundus* antennae, and (2) that eight of these components [all except myrcene and (*E*)-2-nonenal] were essential for the attraction of *M. ridibundus* in behavioral bioassays. A blend of the same 10 components and (+)-limonene (an abundant compound in cocoon volatiles) as an 11th component also attracted/arrested foraging fifth-instar *C. pomonella* larvae (Jumean et al., 2004b). Here we report that a blend of (*E*)-2-octenal, (*E*)-2-nonenal, sulcatone, and geranylacetone in combination with either 3-carene and/or three saturated aldehydes (octanal, nonanal, decanal) elicited attraction or arrestment of pupation site-seeking fifth-instar *C. pomonella* larvae.

METHODS AND MATERIALS

Experimental Insects. Larvae were shipped in trays of artificial diet from the Sterile Insect Release Program rearing facility in Osoyoos, British Columbia, Canada. Trays containing 1000 larvae were kept in a glass aquarium ($60 \times 31 \times 31$ cm) and stored at 15°C under a 16L:8D photoperiod. Nondiapausing fifth-instar larvae were removed from the diet as needed for experiments.

Acquisition of Volatiles. To acquire naturally emitted volatiles for olfactometer experiments, 300 cocoon-spinning male and female fifth-instars (1:1 sex ratio) were placed in a cylindrical Pyrex glass chamber (15.5×20 cm). An empty chamber served as control. A water aspirator drew charcoal-filtered

air at ~2 l/min through each chamber and through a glass column (14 × 1.3 cm OD) containing Porapak Q (50Y80 mesh; Waters Associates, Inc., Milford, MA, USA). After 72 hr, volatiles were eluted from the Porapak Q trap with 3 ml of pentane and ether (95:5). Extracts were concentrated under a nitrogen stream so that 1 μl was equivalent to 10 cocoon-spinning larvae-hour equivalents (10 CSLHE = volatiles released from 10 cocoon-spinning *C. pomonella* larvae during 1 hr). Extracts were stored in darkness at −15°C, and analyzed by coupled gas chromatographyYmass spectrometry (GCYMS) in full-scan electron impact mode, using a Varian Saturn 2000 Ion Trap GCY MS fitted with a DB-5 column (30 m × 0.25 mm i.d., J&W Scientific, Folsom, CA, USA).

Olfactometer Experiments. In two-choice Petri dish olfactometers (detailed drawing in Duthie et al., 2003), test stimuli were randomly assigned to one of two 4-ml vials (Table 1), each with a perforated Eppendorf tube to prevent physical contact of experimental larvae with test stimuli. Stimuli were pipetted onto Whatman no. 1 filter paper disks (1 cm diam.), with treatment and control disks receiving the same amount of solvent. For each replicate, one fifth-instar was placed in the center of the olfactometer, and its pupation site was recorded 18Y24 hr later. All experiments were conducted at 21Y26°C in complete darkness.

To determine whether storage of cocoon volatile extract diminished its attractiveness, experiments 1 and 2 tested the responses of larvae to 180 CSLHE of fresh (1 d old) and aged (8 d old) extracts. In experiment 3, a synthetic blend (SB) of 11 cocoon volatiles [heptanal, octanal, nonanal, decanal, (*E*)-2-octenal, (*E*)-2-nonenal, (+)-limonene, myrcene, 3-carene, sulcatone, geranylacetone] (Jumean et al., 2004b) was tested at 200 CSLHE to determine whether it had a comparable behavioral effect as Porapak Q extract of natural cocoon volatiles. Testing natural vs. synthetic cocoon volatiles, or two blends of synthetic cocoon volatiles, in the same confined olfactometer was attempted but found to compromise the larva's ability to discriminate between volatile blends (Z. Jumean, unpublished data).

Taking the results of experiment 3 into account, experiments 4Y7 tested SB at four doses (1, 10, 100, and 1000 CSLHE) to determine the optimal dose for subsequent experiments. To determine essential pheromone components, experiments 8Y16 tested SB vs. blends lacking certain classes of organic chemicals (Byers, 1992), such as ketones (experiment 8), monoterpenes (experiment 9), or aldehydes (experiment 10). Experiments 11Y16 took a similar approach by deleting from SB either saturated aldehydes (experiment 11), unsaturated aldehydes (experiment 12), or individual components (experiments 13Y16). Considering the results of experiments 8Y16, experiments 17Y26 tested a four-component rudimentary synthetic blend (RSB) [(*E*)-2-nonenal, (*E*)-2-octenal, sulcatone, geranylacetone] alone (experiment 17) or in combination with one of

TABLE 1. DETAILS ON EXPERIMENTAL INSECTS AND STIMULI TESTED IN LABORATORY OLFACTOMETER AND ON-TREE EXPERIMENTS

Petri dish olfactometer bioassays			
Experiment no.	Vial 1	Vial 2	*N*
1	Porapak Q extract (180 CSLHE,[a] 1 d old)	Solvent[b]	30
2	Porapak Q extract (180 CSLHE, 8 d old)	Solvent	32
3	200 Synthetic Blend[c] (SB)	Solvent	35
4	1 SB	Solvent	30
5	10 SB	Solvent	28
6	100 SB	Solvent	31
7	1000 SB	Solvent	26
8	200 SB *minus* ketones	Solvent	35
9	200 SB *minus* monoterpenes	Solvent	35
10	200 SB *minus* aldehydes	Solvent	31
11	200 SB *minus* saturated aldehydes	Solvent	33
12	200 SB *minus* unsaturated aldehydes	Solvent	31
13	200 SB *minus* (*E*)-2-nonenal	Solvent	30
14	200 SB *minus* (*E*)-2-octenal	Solvent	30
15	200 SB *minus* sulcatone	Solvent	33
16	200 SB *minus* geranylacetone	Solvent	33
17	200 Rudimentary Synthetic Blend[d] (RSB)	Solvent	25
18	200 RSB *plus* (+)-limonene	Solvent	20
19	200 RSB *plus* myrcene	Solvent	30
20	200 RSB *plus* 3-carene	Solvent	32
21	200 RSB *plus* heptanal *plus* octanal *plus* nonanal	Solvent	28
22	200 RSB *plus* octanal *plus* nonanal *plus* decanal	Solvent	27
23	200 RSB *plus* heptanal *plus* nonanal *plus* decanal	Solvent	23
24	200 RSB *plus* heptanal *plus* octanal *plus* decanal	Solvent	25
25	200 RSB *plus* octanal *plus* nonanal	Solvent	25
26	200 RSB plus 3-carene *plus* octanal *plus* nonanal *plus* decanal	Solvent	30
27	10 SB	Solvent	40
28	10 SB (3-carene 10-fold increased)	Solvent	33
29	10 SB [(*E*)-2-octenal 10-fold increased]	Solvent	47
30	10 SB [(*E*)-2-nonenal 10-fold increased]	Solvent	39
31	10 SB [(*E*)-2-octenal 10-fold increased plus (*E*)-2-nonenal 10-fold increased]	Solvent	36
On-tree experiments			
	Treatment	Control	
32	CB + 25 9 larvae (1 d old)[e,f]	CB	12
33	1000 SB	Solvent	18
34	100 SB	Solvent	18
35	10,000 SB	Solvent	12
36	100 SB [(*E*)-2-octenal 10-fold increased plus (*E*)-2-nonenal 10-fold increased]	Solvent	12

three monoterpenes [(+)-limonene, myrcene, or 3-carene; experiments 18Y20], or with one of four 3-component blends of saturated aldehydes [heptanal, octanal, nonanal, decanal; experiments 21Y24]. Considering that only the blend of saturated aldehydes that contained octanal, nonanal, and decanal enhanced the effectiveness of RSB (experiment 22), experiment 25 explored whether decanal could be deleted from this blend without affecting the blend's behavioral activity. Experiment 26 then was designed to confirm that the RSB plus four essential components [3-carene (experiment 20); octanal, nonanal, and decanal (experiments 22, 25)] was attractive to larvae seeking pupation sites. Final laboratory experiments 27Y31 explored whether SB at the low and behaviorally inactive dose of 10 CSLHE would become stimulatory upon increasing the amount of 3-carene (experiment 28), or either one or both of (*E*)-2-octenal and (*E*)-2-nonenal (experiments 28Y31).

The on-tree experiments (32Y36) were conducted at Simon Fraser University (May to October 2003) and employed 4-cm wide corrugated cardboard bands (cut from stock of 0.46 × 76 m single-face corrugated cardboard; Shippers Supply Inc., British Columbia, Canada). Cardboard bands were affixed with metal wire 45 cm above ground to trunks of maple (*Acer* spp.) trees that were 10 Y16 cm in diam at that height. Bands were divided into two halves, with test stimuli applied to the waxed center (4 cm^2) of each half.

For each replicate in experiments 32Y36, 20 fifth-instars were released from a thin circular collar affixed to the tree's main branch crotch (~1.50 m above ground). Experiments were started at 22:00 hr and terminated 10Y12 hr later by recording the number of larvae cocooning in treatment or control halves of the cardboard bands.

Experiments 32 and 33 tested whether cardboard band halves baited with 25 1-d-old *C. pomonella* cocoons containing larvae or prepupae (experiment 32), or baited with a synthetic blend at 1000 CSLHE (experiment 33), attracted or arrested more *C. pomonella* larvae than did unbaited cardboard band halves. In

Footnotes to Table 1

[a] CSLHE = cocoon-spinning larvae hour equivalents.

[b] Solvent consisted of redistilled pentane (20 Y100 μl).

[c] 10 SB = synthetic blend of 11 components: decanal (1.4 ng) [Aldrich], nonanal (4.1 ng) [Aldrich], octanal (0.94 ng) [Aldrich], heptanal (0.85 ng) [Aldrich], (*E*)-2-nonenal (1.00 ng) [Bedoukian], (*E*)-2-octenal (0.41 ng) [Bedoukian], geranylacetone (0.50 ng) [Aldrich], sulcatone (0.81 ng) [Aldrich], 3-carene (0.95 ng) [Aldrich], myrcene (0.84 ng) [Aldrich], (+)-limonene (10.00 ng) [Aldrich].

[d] 10 RSB = rudimentary synthetic blend of four components: (*E*)-2-nonenal (1.00 ng), (*E*)-2-octenal (0.41 ng), geranylacetone (0.50 ng), sulcatone (0.81 ng).

[e] Fifth-instar larvae were allowed to cocoon in an open-fluted cardboard (CB) strip (4 cm^2).

[f] Female larvae were used as test stimuli, and male larvae were bioassayed to allow recognition and recording of all those larvae that had responded to test stimuli.

experiment 32, female larvae served as test stimuli and male larvae (as determined by testes visible through the dorsal integument) were bioassayed to allow recognition and recording of those larvae that had responded to test stimuli. This experimental design was justified because both male and female cocoon-spinning larvae produce and respond to the same volatile components (Jumean et al., 2004a). Experiments 34Y35 determined whether a 10-fold decrease (experiment 34) or a 10-fold increase (experiment 35) in the dose of the synthetic blend affected the response of *C. pomonella* larvae in the field. Finally, experiment 36 explored whether the synthetic blend at the low and behaviorally inactive dose of 100 CSLHE (see experiment 34) would become active by increasing the amounts of (*E*)-2-octenal and (*E*)-2-nonenal as essential blend components.

Statistical Analyses. Numbers of larvae responding to treatment and control stimuli in laboratory olfactometer experiments were analyzed with the χ^2 goodness-of-fit test, using Yates correction for continuity ($\alpha = 0.05$) (Zar, 1999). Numbers of larvae responding to treatment and control stimuli in on-tree bioassays were analyzed with the Wilcoxon paired-sample test ($\alpha = 0.05$) (Zar, 1999).

RESULTS

In laboratory olfactometer experiments, both fresh and aged Porapak extracts of cocoon volatiles at 180 CSLHE attracted larvae (Figure 1; experiments 1, 2), as did a synthetic blend (SB) of 11 candidate pheromone components at 200 CSLHE (Figure 1, experiment 3). SB elicited a behavioral response also at 100 CSLHE (Figure 1; experiment 6), but not at 1, 10, or 1000 CSLHE (Figure 1; experiments 4, 5, 7). SBs lacking ketone or monoterpene components remained moderately attractive (Figure 2; experiments 8, 9), whereas an SB lacking aldehydes was inactive (Figure 2; experiment 10). An SB lacking saturated aldehydes was still active (Figure 2; experiment 11), whereas SBs lacking one or both unsaturated aldehydes [(*E*)-2-nonenal or (*E*)-octenal], or ketones [sulcatone or geranylacetone] elicited no significant responses from larvae (Figure 2; experiments 12Y16). A rudimentary synthetic blend [RSB: (*E*)-2-octenal, (*E*)-2-nonenal, sulcatone, and geranylacetone] was not attractive (Figure 3; experiment 17), but the addition of 3-carene, unlike myrcene or (+)-limonene, rendered RSB attractive (Figure 3; experiments 18Y20). Addition of the three saturated aldehydes octanal, nonanal, and decanal, unlike other three-component blends of saturated aldehydes, also rendered RSB attractive (Figure 3; experiments 21Y24) to a level comparable with that of RSB plus all four saturated aldehydes (= SB minus monoterpenes; experiment 9).

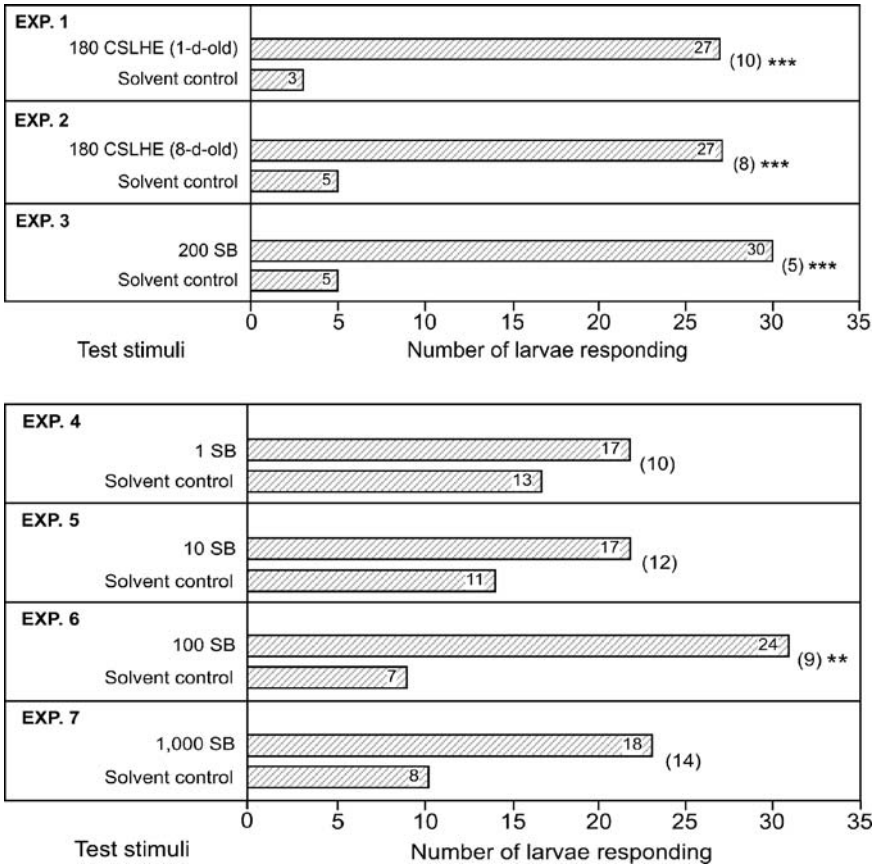
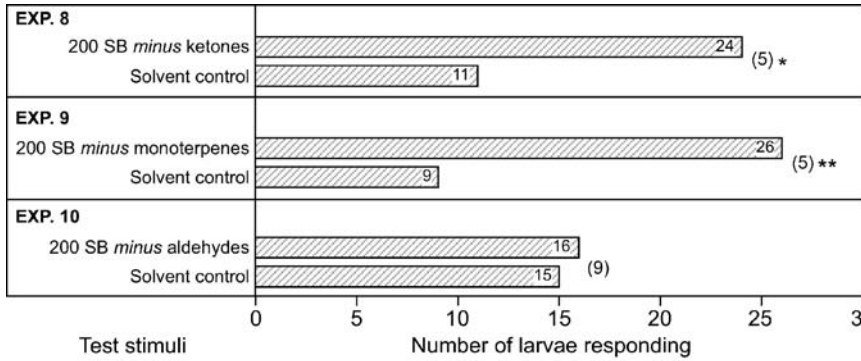


FIG. 1. Response of fifth-instar *Cydia pomonella* larvae in Petri dish olfactometers to extracts of cocoon-derived volatiles and to a synthetic blend (SB) of 11 candidate pheromone components (experiments 1Y3), or to varying doses of SB (experiments 4Y7). Number of larvae responding to each stimulus is given within bars; number of larvae not responding in each experiment given within parentheses; asterisks indicate a significant response to a particular treatment; χ^2 test with Yates correction for continuity; $^{**}P < 0.005$; $^{***}P < 0.001$. Ten SB consisted of three monoterpenes [(+)-limonene (10.00 ng), 3-carene (0.95 ng), myrcene (0.84 ng)], four saturated aldehydes (heptanal, octanal, nonanal, decanal), two unsaturated aldehydes [(*E*)-2-octenal (0.41 ng), (*E*)-2-nonenal (1.00 ng)], and two ketones [sulcatone (0.81 ng), geranylacetone (0.50 ng)]. Cocoons were 1Y3 d old at the time of aeration but Porapak Q extracts were tested before and after aging to determine stability of semiochemicals. Aliquots of 180 or 200 CSLHE (cocoon-spinning larvae hour equivalents) were tested in experiments 1Y3. Aliquots of 1, 10, 100, or 1000 CSLHE were tested in experiments 4Y7; the same amount (20Y25 μl) of pentane was applied to treatment and control stimuli in experiments 1Y7.



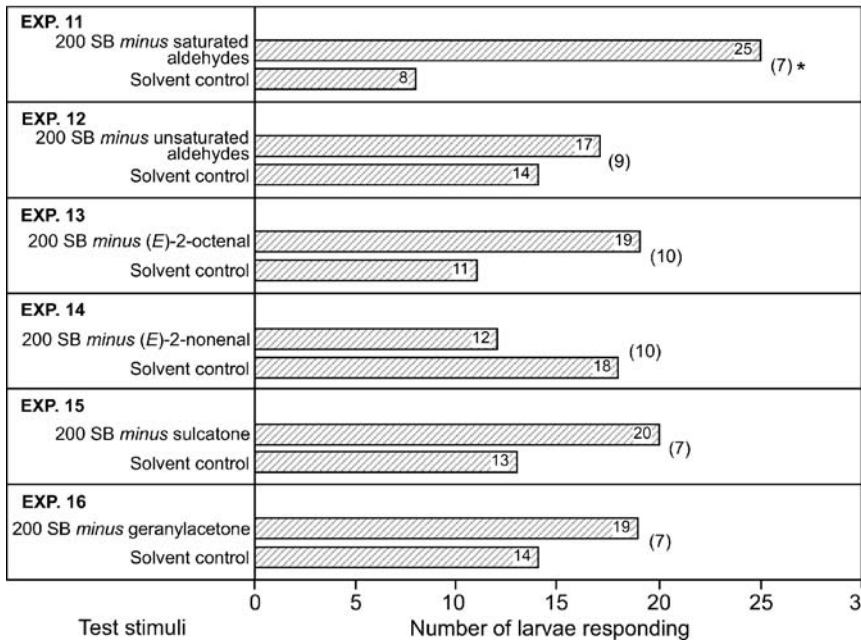


FIG. 2. Response of fifth-instar *Cydia pomonella* larvae in Petri dish olfactometers to synthetic blends (SB) lacking one or more candidate pheromone components. Number of larvae responding to each stimulus is given within bars; number of larvae not responding given in parentheses; asterisks indicate a significant response to a particular treatment; χ^2 test with Yates correction for continuity; $^*P < 0.05$; $^{**}P < 0.01$. Aliquots of 200 CSLHE (see caption of Figure 1) were tested. The same amount (20 µl) of pentane was applied to treatment and control stimuli.

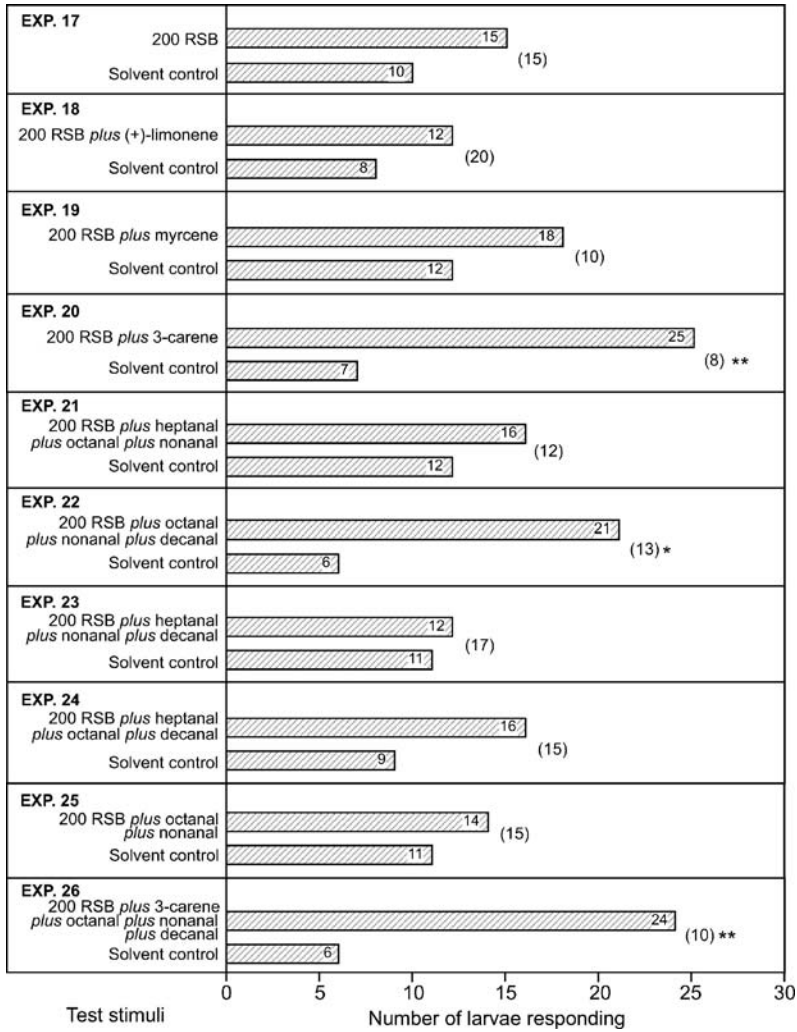


FIG. 3. Response of fifth-instar *Cydia pomonella* larvae in Petri dish olfactometer experiments 17Y26 to a rudimentary synthetic blend (RSB) of pheromone components and to RSB plus individual or groups of candidate pheromone components. Number of larvae responding to each stimulus is given within bars; number of larvae not responding given in parentheses; asterisks indicate a significant response to a particular treatment; χ^2 test with Yates correction for continuity; $^*P < 0.01$; $^{**}P < 0.005$. Ten RSB consisted of two unsaturated aldehydes [(*E*)-2-octenal (0.41 ng), (*E*)-2-nonenal (1.00 ng)] and two ketones [sulcatone (0.81 ng), geranylacetone (0.50 ng)]. Aliquots of 200 CSLHE (see caption of Figure 1) were tested. The same amount (20 μl) of pentane was applied to treatment and control stimuli.

Addition of only octanal and nonanal to RSB failed to elicit a response from pupation site-seeking larvae (Figure 3; experiment 25), but RSB plus 3-carene, and octanal, nonanal, and decanal did elicit a behavioral response (Figure 3; experiment 26). SB at the low dose of 10 CSLHE had no effect on larval behavior (Figure 4; experiment 27). Ten-fold increases of either 3-carene, (*E*)-2-octenal, or (*E*)-2-nonenal in that low-dose blend did not modify its attractiveness (Figure 4; experiments 28Y30) but a 10-fold increase of both (*E*)-2-octenal and (*E*)-2-nonenal in that blend stimulated a positive response from larvae (Figure 4; experiment 31).

In on-tree experiments, cocoons from conspecifics (experiment 32), and SB at 1000 CSLHE (experiment 33), attracted or arrested *C. pomonella* larvae foraging for pupation sites (Figure 5). In contrast, SB at 100 or 10,000 CSLHE was not active (Figure 5; experiments 34Y35). Although the SB had no behavioral effect at 100 CSLHE (experiment 34), a 10-fold increase of both (*E*)-

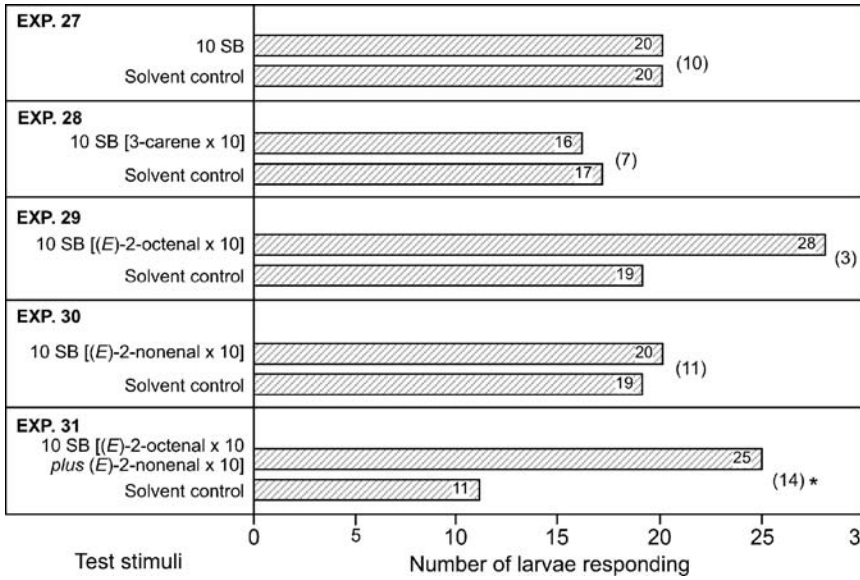
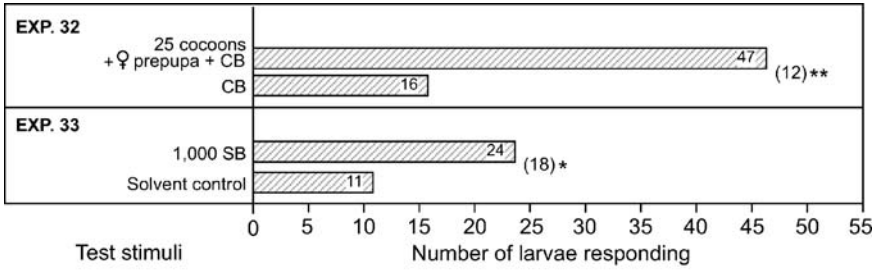
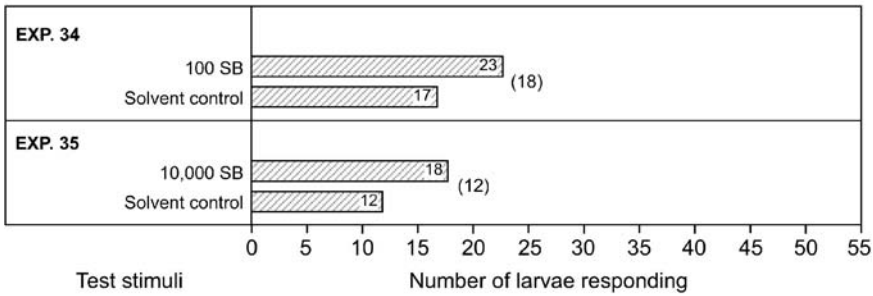


FIG. 4. Response of fifth-instar *Cydia pomonella* larvae in Petri dish olfactometer experiments 27Y31 to a synthetic blend (SB; see caption of Figure 1) of 11 components with the relative proportion of 3-carene or unsaturated aldehydes [(*E*)-2-octenal, (*E*)-2-nonenal] increased by 10-fold. Number of larvae responding to each stimulus is given within bars; number of larvae not responding given in parentheses; asterisks indicate a significant response to a particular treatment; χ^2 test with Yates correction for continuity; $^*P < 0.05$. Aliquots of 10 CSLHE (see caption of Figure 1) were tested. The same amount (20 μl) of pentane was applied to treatment and control stimuli.





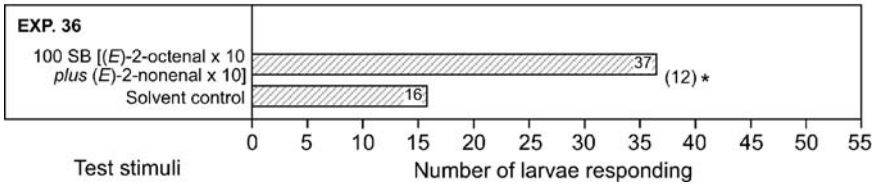


FIG. 5. Response of male (experiment 32), or male and female (experiments 33Ÿ36), fifth-instar *Cydia pomonella* larvae in on-tree experiments to stimuli consisting of either 25 cocoons with female larvae (experiment 32), or synthetic blends (SB) of 11 components (see caption of Figure 1) at varying doses and component ratios. Strips of corrugated cardboard (CB) served as a pupation site. Number of larvae responding to each stimulus is given within bars; number of replicates given in parentheses; asterisks indicate a significant response to a particular treatment; Wilcoxon paired-sample test; $*P < 0.01$; $**P < 0.005$. Aliquots of 100, 1,000, or 10,000 CSLHE (see caption of Figure 1) were tested. The same amount (100 µl) of pentane was applied to treatment and control stimuli. In experiment 36 the proportion of (*E*)-2-octenal and (*E*)-2-nonenal in the blend was increased by 10-fold.

2-octenal and (*E*)-2-nonenal in that blend produced responses from larvae (experiment 36).

DISCUSSION

Our laboratory and field data provide evidence that cocoon-spinning *C. pomonella* larvae produce a pheromone that attracts and/or arrests conspecific larvae seeking pupation sites (Duthie et al., 2003; Jumean et al., 2004a).

The cocoon-derived 11 candidate pheromone components that were bioassayed in olfactometer experiments 1Y31 were selected based on evidence that they attracted not only *M. ridibundus* parasitoids but also *C. pomonella* larvae (Jumean et al., 2004b). The comparable biological activity of the 11-component synthetic blend and the Porapak Q extract of cocoon volatiles (Figure 1, experiments 1Y3) suggested that all essential pheromone components were present in the synthetic blend.

The aggregation pheromone of *C. pomonella* larvae is surprisingly complex and responses were critically dependent on dose and blend composition. Low- or high-dose blends were ineffective (Figure 1), as were blends lacking either (*E*)-2-octenal, (*E*)-2-nonenal, sulcatone, or geranylacetone (Figure 2; experiments 13Y16). Synergism between components was also evident when the four-component rudimentary blend of (*E*)-2-octenal, (*E*)-2-nonenal, sulcatone, and geranylacetone failed to affect larval behavior (Figure 3, experiment 17), but addition of either 3-carene (experiment 20) or three saturated aldehydes (octanal, nonanal, decanal) (experiment 23) resulted in attractive blends. Similar positive effects caused by 3-carene, or by saturated aldehydes, suggested redundancy in the blend composition. With five components needed for *C. pomonella* larvae to respond (Figure 3; experiment 20), and eight components needed for *M. ridibundus* parasitoids to respond (Jumean et al., 2004b), it appears that *M. ridibundus* requires a more complex signal to locate *C. pomonella* host prepupae than *C. pomonella* larvae require to communicate among themselves. The fact that the pheromone at low dose but with 10-fold increase of (*E*)-2-octenal and (*E*)-2-nonenal elicited a behavioral response from larvae (Figure 4; experiment 31) suggests that both of these unsaturated aldehydes are major components of the pheromone blend.

Pheromone components are perceived by *C. pomonella* larvae as airborne signals, because baffles in olfactometer experiments prevented physical contact with natural or synthetic pheromone. However, whether the pheromone serves primarily to attract or arrest conspecific larvae is not yet known.

The possible adaptive significance of pheromone-based larval aggregation will be intriguing to investigate. Duthie et al. (2003) proposed that

aggregations of *C. pomonella* larvae are part of a reproductive strategy that facilitates the earliest possible mating of eclosed adults. The proposed fitness advantage, however, may be offset by costs associated with larval aggregations. Host-derived pheromones are reliable indicators of host presence (Wiskerke et al., 1993; Wertheim et al., 2003), and are exploited by foraging parasitoids as illicit receivers of such signals (Stowe et al., 1995; Haynes and Yeargan, 1999; Jumean et al., 2004b). If, however, foraging parasitoids are egg-limited (Bezemer and Mills, 2001), individual *C. pomonella* larvae or prepupae in aggregations may be at a lower risk of parasitism than larvae that cocoon in isolation.

Aggregation of *C. pomonella* larvae as part of a proposed reproductive strategy (Duthie et al., 2003) might explain localized fruit damage in orchards treated with synthetic sex pheromone for *C. pomonella* control. Female *C. pomonella* eclosing from larval aggregations might be mated irrespective of otherwise functional pheromone-based tactics to disorient or attract and kill mate-foraging males. With the larval aggregation pheromone identified in this study, and shown to attract or arrest larvae in on-tree experiments (Figure 5; experiments 32Y36), there may be a new opportunity to manipulate *C. pomonella* larvae in commercial fruit or nut orchards. Larval manipulation would be compatible with other biorational tactics of *C. pomonella* control including pheromone-mediated mating disruption and postharvest fruit removal (Judd et al., 1997).

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